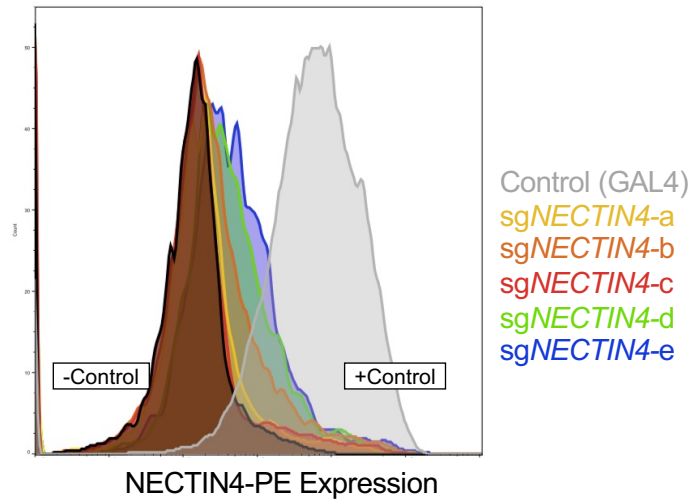
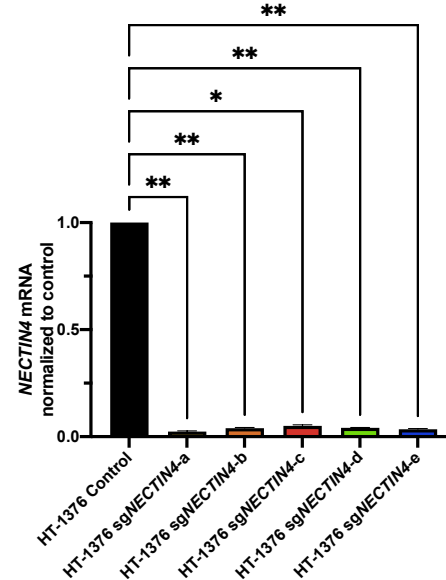
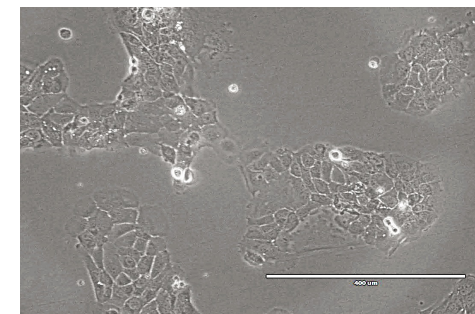
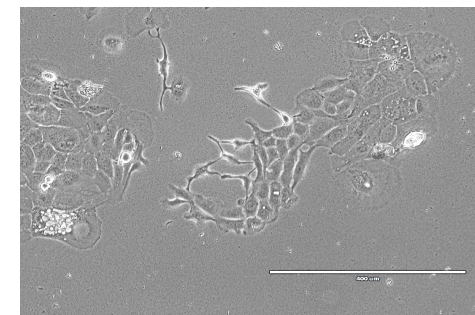
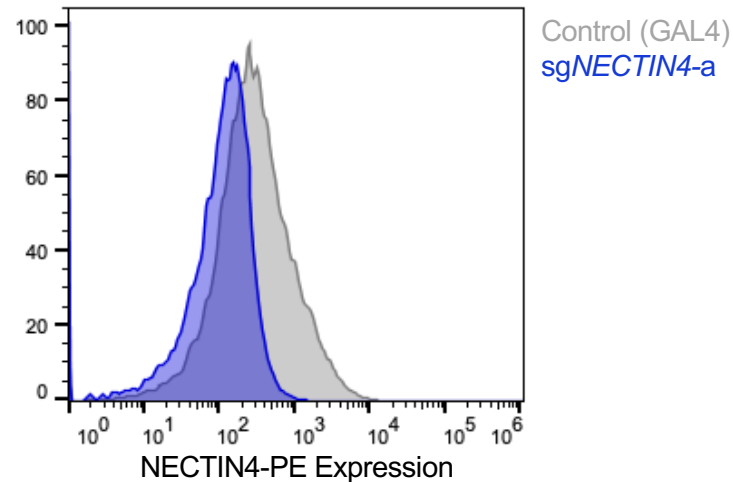
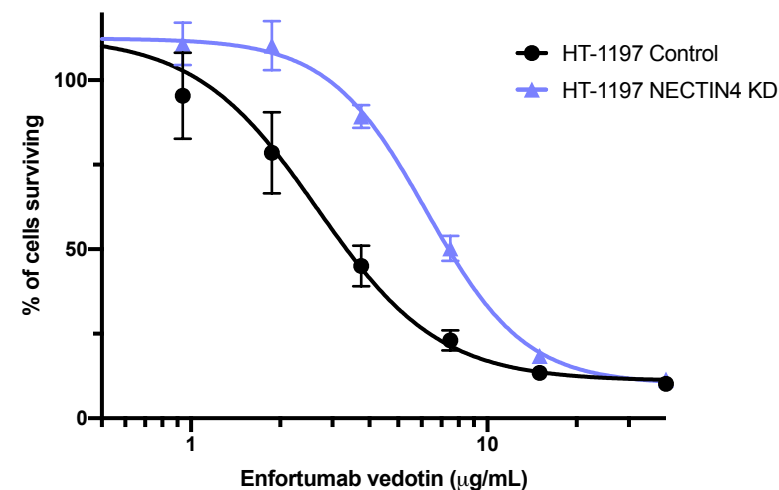


A**HT-1376 NECTIN4 knockdown****B****C**

HT-1376 Control



HT-1376 NECTIN4 Knockdown

D**HT-1197 NECTIN4 knockdown****E**

Supplementary Figure 8. (A) Validation of knockdown by cell surface staining of NECTIN4 protein expression in HT-1376 cells using CRISPRi/dCas9 and five unique sgRNAs targeting different regions of *NECTIN4* gene and promoter. The sgNECTIN4-a guide was used for subsequent experiments. (B) Confirmation of sgNECTIN4 CRISPRi/dCas9-mediated *NECTIN4* mRNA knockdown in HT-1376 cells by quantitative PCR, ** $p < 0.01$. All sgRNAs decreased *NECTIN4* mRNA expression by >90%. (C) Phase-contrast, brightfield microscopy of HT-1376 cells in Control and NECTIN4 knockdown cells to show the morphology of cells. Scale bar = 400 μm . (D) Validation of knockdown by cell surface staining of NECTIN4 protein expression in HT-1197 cells using CRISPRi/dCas9. (E) EV dose-response WST proliferation assay in HT-1197 Control (black) and NECTIN4 (KD) knockdown (purple) cells.